

POPULATION STRUCTURE OF XYLELLA FASTIDIOSA OF CITRUS VARIEGATED CHLOROSIS IN SAO PAULO STATE: FROM A SMALL BRANCH TO THE WHOLE STATE

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ABSTRACT

The economic and social importance of citrus production in Brazil and the high economic loss caused by citrus variegated chlorosis (CVC) in Brazilian orchards have resulted in an extensive research program involving the sequencing of the genome of *Xylella fastidiosa* (*Xf*) subsp. *pauca* (*Xf-pa*, strain 9a5c), the causal agent of CVC. Other genomes of *Xf-pa* are currently being sequenced. Despite the great interest in genomics of *Xf-pa*, relatively little information is available about the population genetics of this pathogen. We previously showed that different *Xf-pa* haplotypes were found in CVC-diseased plant and hypothesized that it was a consequence of multiple inoculation events originating from different sources (Coletta-Filho and Machado, 2003, Geographical genetic structure of *Xylella fastidiosa* from citrus in São Paulo State, Brazil Phytopathology, 93:28-34). Based on those findings we studied the role of spatial scale on the genetic diversity of *Xf-pa* in sweet orange plants affected by CVC from various geographical regions of Sao Paulo state. We used analyses of molecular variance (AMOVA) to test hypothesis that genetic differentiation among populations increased from central (branch) to peripheral sites (geographic regions). Spatial scale ranged from 4–6 cm long branches to hundreds of kilometers, including within and among citrus plant comparisons. At the smallest spatial scale (single branch) five isolates were obtained, 10 to 15 isolates were obtained from each plant, and 40 isolates total from each orchard (geographic regions), in total 160 isolates from four geographical regions were typed. The genetic structure of *Xf-pa* was determined using 16 simple sequence repeats (SSR) loci. AMOVA analysis using the Euclidean squared distances between pairs of isolate) revealed significant genetic differentiation among all population levels: i) within branches ($F_{ST} = 0.856$, Var. = 14.0%); ii) among branches within plants ($F_{SC} = 0.374$, Var. = 8.58%); iii) among plants within regions ($F_{SC} = 0.77$, Var. = 40.75%); and iv) among regions ($F_{CT} = 0.44$, Var. = 36.71%). These results indicate that spread of CVC in Sao Paulo state occurred in a wave-like fashion, but a significant percentage of genetic variability among isolates was observed at the smallest scale sampled (small branches), which could be attributed to natural recombination events among isolates rather than different sources of multiple infections, as previously hypothesized. On the other hand, it is possible that the loci used here evolve quickly, which would also be a possible interpretation for these data. Future work is needed to address these competing hypotheses.

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